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A FURTHER ANALYSIS OF THE HEREDITARY TRANSMISSION OF DEGENERACY AND DEFORMITIES BY THE DESCENDANTS OF ALCOHOLIZED MAMMALS

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INTRODUCTION

A LITTLE more than two years ago the senior author (Stockard, '13) recorded in this journal experiments which had then been running for three years and seemed to show a definite injury of the germ cells by treating mammals with the fumes of alcohol. This injury of the male germ cells is of such a nature that an alcoholized male guinea pig almost invariably begets defective offspring even when mated with a vigorous normal female. At that time it was also shown that F_1 animals, the offspring of treated parents, though themselves not treated,

had the power to transmit the defective condition to their young, and such F_2 young were equally if not more defective than the immediate offspring of the treated animals.

In 1914 in a short abstract Stockard showed further that the offspring from F_2 individuals were apparently more defective than their parents and were often grossly deformed. One case was recorded of the occurrence of a litter of two F_3 animals, both of which were extremely weak and neurotic, showing a condition suggesting paralysis agitans, and further than this the two animals were typical anophthalmic monsters. The eyes were completely absent, no optic nerve or optic chiasma or visible optic tracts along the tuber cinereum could be found on a careful gross examination of the brain. The two animals were produced by parents (F_2) that had never been treated with alcohol, the four grandparents (F_1) had also not been treated, while the *three great grandfathers* had been alcoholized and the three great grandmothers were normal untreated individuals.

Defective eyes and absence of one eye or both eyes have been frequently met with in the experiment, as well as the peculiar nervous condition, and these symptoms are to be considered indicative of the injury or change induced in the male germ cells by the experimental treatment, which in the above case was transmitted through three generations. No question could remain as to the action on the germ cells, as only male ancestors had been treated; every female of the line was an untreated animal.

This abstract called attention to the fact that there was a tendency for the results to differ in subsequent generations from treated males as compared with the descendants of treated females—not enough data were then present to offer any explanation of these differences and a consideration of them will be undertaken in the present paper.

At that stage of the experiment it was also difficult to offer an exact analysis of the mode of transmission of

the defects and the type of injury induced by the alcohol treatment, since the total numbers were not large and the F_2 animals had only a few matings, while further generations had not become available for breeding.

The same experiments have now been continued for more than five years and a number of animals have been used, over 700, which cover the behavior of four generations and supply data of sufficient extent to allow a more thorough analytical consideration of the heredity problem concerned.

Experiments of this nature on mammals are fraught with many difficulties, slowness of breeding, small size of litters, difficulty of handling, etc. Yet such material offers one very great advantage in that the quality of the offspring and generations studied is of such a complex that one is enabled to detect indications of rather slight injuries or changes in the material carriers of heredity which would not become evident on lower forms with less diversity in their methods of behavior and structural appearance. In other words, we take it that such conditions as are spoken of as racial degeneracy in man and mammals are often very difficult or even at times impossible to detect in lower forms.

These conditions are for many reasons thought to be inherited. If so their inheritance must be due to a pathological condition of the material carriers of heredity, the chromosomes, or what not, since they are not normal states and, like diseases, are constantly arising in normal families on account of one or another form of intoxication. Is it possible then to produce such a racial degeneracy artificially by treating only one generation of the animals and by so doing observe a pathological behavior of the carriers of heredity? Arguing from analogy there must be pathological heredity due to diseased or altered chromosomes in the germ cells just as truly as there is a known pathological behavior of every other organ and tissue of the animal body.

It becomes then a problem to study the possible meth-

ods of modifying the chromosomes or carriers of the inherited qualities of organisms in order to further analyze their normal physiological behavior; in the same way that experimental embryology has been able to supply so many valuable clues to the normal processes of development.

In the following pages we believe the facts indicate that individual guinea pigs are now living in this experiment that have had the carriers of hereditary qualities, the chromatin, of their germ cells injured for a longer time than four years. And during this time they have given rise to offspring of more or less degenerate or deformed type, and in some cases these offspring have passed this modified chromatin on through three generations, all of which contain pathological chromatin and show somatic defects and deformities as an index of their tainted chromatic ancestry. Modified chromatin has been living in the experiment for more than four years in five different generations of animals as a result of the treatment on the one original, P_1, parent generation.

We have tried to regulate every controllable source of error and there can be no doubt that the conditions are brought about in the way described. Could the degeneracy which is so pronounced have previously existed in the stock? This question has been controlled in the first place by the use of two entirely different stocks from different sources and obtained one and one half years apart, the first in the fall of 1910 and the other in the early winter of 1912. The responses of the two stocks to the experimental treatment have been identical. As a second method of control every animal has been tested by one or more normal matings before being introduced into the experiment, and only those giving normally strong offspring have been used. A further crucial control is the constant mating of normal untreated animals from both stocks under identical cage conditions with the experimental individuals. These animals continue to breed normally until very old, when they gradually become

sterile. But none have ever given rise to a defective or deformed individual, and the rate of mortality of the young indicates the average healthy condition found in normal guinea-pig breeding. There is a striking contrast between the records of these normal young and the mortality record, the frequency of easily recognized nervous symptoms of degeneracy, and the prevalence of gross deformities in the experimental races.

The external as well as internal factors are to be considered not only in individual or embryonic development, but also in heredity. And the present experiments now demonstrate for mammals that either the spermatozoon or the ovum may be experimentally injured or modified in such a manner as not only to give rise to (abnormal) subnormal development in the resulting embryo, but the effects of the injury may be transmitted from generation to generation, until an affected line actually fades out through degeneracy and sterility as a result of the transmitted condition.

Material and Methods

The animals used in the experiments have been ordinary vigorous guinea pigs of large size, particular care being taken to select animals less than one year old to begin with and good breeders.

At the beginning of the experiments alcohol was given along with the food, but the animals ate less and the food usually disagreed with them. It was then administered in diluted form by a stomach tube; this method was even more unsuccessful, disturbing digestion and seeming to upset the animals considerably. It is certain that alcohol given to animals through the stomach deranges their appetite and digestion to such an extent that the experimenter is unable to determine whether the resulting effects are due to the alcohol, as such, or to the generally deranged metabolism of the animal. When given in drinking water they take little or none of the water and the treatment is insufficient. For these reasons an inha-

lation method of treatment was resorted to early in the study, and, as far as experience goes, it has no serious disadvantages and does not complicate the conditions of the experiment.

This method may be merely described in brief for the convenience of the reader, since it has been fully recorded with illustrations of the fume tanks in previous publications. A fume tank of copper is made of sufficient size to supply breathing space for four or five guinea pigs at one time. The tank has four outlets, so that a definite amount of fumes may be passed through in a given time and the ventilation controlled. In this way each animal could be given a definite measured dose. The individuals, however, differ so much in their resistance to the treatment that it has been found better to treat all to about the same degree of intoxication. Such a physiological index is more reliable, since every animal may be affected to the same degree each day. For this purpose the animals are placed in the fume tank on a wire screen, and absorbent cotton soaked with alcohol is placed beneath the screen, so that they inhale the alcohol fumes arising from the cotton to saturate the atmosphere of the tank.

Ether was given in a similar manner. The animals are much more readily overcome by these fumes and must be carefully watched while inhaling even the most dilute doses.

To avoid handling the females during pregnancy, special treating cages are devised. An ordinary box-run with a covered nest in which the animal lives is connected by a drop-door with a metal-lined tank, having a similar screen arrangement, etc., to that of the general treatment tank. The pregnant animal may be driven daily into the tank and thus treated with alcohol fumes throughout her pregnancy without being handled in any way that might disturb the developing fetus.

Particular care is necessary in mating the animals in such an experiment, as the females are often slow to con-

ceive and some of the F_2 and F_3 individuals of both sexes are not very prolific and in many cases are almost or quite sterile. Each female is kept in a separate run and the male is placed in with her just before the time of the expected heat period, ovulation, and he remains in her cage for from two to three weeks so as to be present at the second ovulation, provided the female was not made pregnant by the first mating. The ovarian cycle of the guinea pig as worked out by L. Loeb seems to correspond closely to what is found in mating experiences.

After mating, the male is removed from the cage and the female remains alone until the young are born. These are left with the mother for about fifteen days, then separated, and the female mated again. In this way the normal females may sometimes give as many as four litters per year, but the experimental animals breed much slower and it is difficult to get even three litters per year.

Direct Effects of the Alcohol Treatment on the Animals

Several of the guinea pigs have now been treated with the fumes of alcohol almost to the point of intoxication for six days per week for a period of five years. This is a considerable space in the life of a guinea pig, which probably would not often extend beyond six or seven years.

The animals are affected by the alcohol fumes in various ways; some of them are stupefied and become drowsy, while others become stimulated and excited and sometimes even vicious, constantly fighting and biting at the other animals in the fume tank. The fumes inhaled into the lungs pass directly into the circulation, so that the animals show signs of intoxication very soon after being put into the tank, yet the intake of alcohol is so gradual that they may remain for one hour or more without becoming totally anesthetized.

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irritated during the early stages of the treatment, but develops a resistance so that later little effect can be noticed. The cornea of the eye is greatly irritated, often becoming milky white and opaque during the first few months. In some cases this later clears and the animal is again able to see, though some of the animals treated for several years have remained entirely blind. The general condition of the animals under the fume treatment is very good. They all continue to grow if the treatment is begun before reaching their full size, and become fat and vigorous, taking plenty of food and behaving in a typically normal manner.

Some of the treated animals have died and others have been killed at different times during the progress of the experiment and their organs and tissues examined carefully and then studied microscopically. All have seemed practically normal. Tissues from several animals treated as long as three years have been examined and the heart, stomach, lungs, kidneys, and other organs present no noticeable conditions that might not be found in normal individuals. Alcoholized animals are usually fat, but there is no fatty accumulation in the parenchyma of any of the organs.

Several of the animals, both males and females, have been partially castrated during the experiments and the ovaries and testes have been found to be in a healthy condition, though certain possible changes may be present which are now being closely studied cytologically and experimentally.

The treated animals are, therefore, little changed or injured so far as their behavior and structure goes. Nevertheless, the effects of the treatment are most emphatically shown by the type of offspring to which the alcoholized individuals give rise, whether they be mated together or with normal individuals. The further significance of the nature of the effects is indicated by the quality of the subsequent generations descended from such an ancestry.

Influence of the Treatment on the Descendants of Alcoholized Animals

It may be well in the first place to consider the results of the experiments from a general standpoint and then to undertake an analysis of the reactions and conditions presented in the several generations and from the several lineal combinations. The records of the matings of the alcoholized animals in various pairs, the control or normal matings, and the matings of the F_1 and F_2 generations, the children and grandchildren of the alcoholized individuals are summarized in the general Table I. This table gives a record of all the matings of the kinds indicated up to July 1, 1915. A similar table was published two years ago, when the number of animals considered was much smaller and the actual indications from the results were less certain than now. On comparing this table with the former one, however, it will be seen that the continuation of the experiments has fully substantiated the results as previously recorded. The table now shows the records of 571 matings which produced 682 full-term young and 189 early abortions or negative results. These numbers are now of considerable magnitude in spite of the fact that the experiment is conducted on mammals which produce only small litters and breed slowly as compared with lower animal forms.

In the first horizontal line the record of pairing alcoholized male guinea pigs with normal females is given. This combination could only produce defective or subnormal young as a result of the injured male germ cells, since the ova are normal and develop in a normal untreated mother. This then is the definite test of the influence of the alcohol treatment on the germ cells.

Ninety such matings have in 37 cases given negative results; that is, failures to conceive, or early abortions. Thus 41 per cent. of the matings of such males were nonproductive, while less than 25 per cent. of normal matings under the same breeding conditions failed to produce full-term litters. Ten stillborn litters, each consisting of

two young, twenty stillborn young, resulted from the 90 matings. While the 90 control matings gave only two stillborn litters, and in both cases these were unusually large litters of four individuals each, and they were probably dead on account of the fact that the mother could not give normal birth to so many offspring. The stillborn litters by the alcoholized fathers were all ordinary-size litters of two young. Thus, while 11 per cent. of the matings of alcoholized males resulted in stillborn litters, only 2 per cent. stillborn litters occurred from normal matings. Forty-three living litters were produced or a little less than 48 per cent. of the matings gave full-term living young, while 73 per cent. of the normal matings give living litters of young.

The 43 litters from alcoholic fathers contained in all 82 young, and 35, or almost 43 per cent., of these died soon after birth, while 66 similar litters from the control lost only 19 young, or 16 per cent., out of 118 individuals. Finally, then, from the 90 matings of alcoholic males with normal mates only 43 full-term litters resulted, consisting in all of 102 young; 55 of these, or 54 per cent., died at birth or soon after, and only 47 individuals, or 46 per cent., survived. Only about half as good record as the 78.5 per cent. surviving young from the matings of normal animals. Almost all of the offspring were very excitable, nervous animals and three of them showed gross deformities of the eyes, while no such conditions were found among any of the offspring of normal animals bred under identical conditions.

These records leave no doubt that the alcoholized male guinea pig is injured in such a way as to induce a decidedly bad effect upon the quality and mortality of his offspring when compared with the records from normal animals.

The second horizontal line of Table I shows the results obtained when alcoholized female guinea pigs are paired with normal males. In this case there is a double chance to injure the offspring. First through the influence of

the treatment on the oocytes or the unfertilized ovarian egg, a direct effect on the germ cells comparable to the injury of the germ cells in the case of the treated males considered above. While in the second place, the developing embryo in the uterus of an alcoholized female may be directly affected by the strange substances contained in the blood and body fluids of the mother. Thus a defective individual may be produced as a result of development in an unfavorable environment or as a result of being derived from an injured or defective egg cell.

Thirty-three matings of alcoholized females with normal males have in seven cases, 21 per cent., given negative results or early abortions; this compares very favorably with the records of the control animals. Four stillborn litters consisting of three individuals each were produced. This is a record of 12 per cent. stillborn litters against only 2 per cent. from normal matings. The alcoholized females gave birth to 22 living litters containing 44 young, and 23, or 52 per cent., of these died, only 48 per cent. surviving against 84 per cent. survivals among the young of similar control litters. The records of the matings of alcoholized females compare very unfavorably with the record of the control matings. Yet the behavior of the alcoholized females is very little, if any, worse than the records shown by the alcoholized males in spite of the double chance the female has to injure her young.

The third horizontal line of the table indicates the results obtained when alcoholized males are paired with alcoholized females. Here there is every chance for the treatment to show its effect. The percentage of early abortions or negative results is very high, about 50 per cent. more than double that of the control matings. Ten per cent. of the matings produced stillborn litters each consisting of two young. Only 17 living litters were born out of 41 matings, about 41 per cent., against 73 per cent. living litters from 90 control matings. The 17 living lit-

ters contained only 26 young, and 12 of these, or 46 per cent., died soon after birth, while but 16 per cent., or one third as many, of the control offspring died out of a total of 118 individuals. The data from the double alcoholic matings is, therefore, extremely bad in the light of normal matings from the same animal stocks bred under exactly the same cage and food conditions.

The fourth horizontal line summarizes the records of all the matings of directly alcoholized animals. In all 164 such matings have been made; 64 of these, or almost 40 per cent., gave negative results or early abortions. Eighteen stillborn litters occurred, consisting of 40 individuals against only two questionable stillborn litters from 90 control matings. Eighty-two, or only 61 per cent., living litters were born, consisting of 152 individuals, 82, or 54 per cent., of which survived and 70, or 46 per cent., died soon after birth; in all 110 full-term young died, while only 82, or 42 per cent., of the total 192 full-term young resulting from the 164 alcoholic matings survived. On the other hand, out of a total of 126 full-term young from only 90 control matings, 99, or 78.5 per cent., survived. The control matings were far more prolific than those of the alcoholized animals and the condition of the young as indicated by the mortality record was far superior to that of the alcoholic offspring.

The fifth line records the outcome of 90 control matings which have been scattered through the entire progress of the experiment under exactly the same conditions and from the same animal stocks as the experimental matings. Eighty-four per cent. of the young in the 66 living litters resulting from the matings of normal animals have survived and all are strong, healthy individuals; in not one instance do they show an indication of nervous degeneracy or any type of recognizable structural deformity, while such degeneracy as well as deformities are extremely prevalent among the offspring and descendants of the alcoholized animals. One other point to be mentioned in considering the records of the

TABLE I

EFFECTS OF ALCOHOL ON THE DESCENDANTS OF TREATED ANIMALS

Condition of the Animals	Number of Matings	Negative Result of Early Abortion	Stillborn Litters	Number of Stillborn Young	Living Litters	Young Dying Soon After Birth	Total Dead	Surviving Young
Alcoholic ♂ ×norm. ♀	90	37	10	20	43	35, 1 c.e.	55	47, 2 c.e.
Norm. ♂ ×alcoholic ♀	33	7	4	12	22	23	35	21
Alcoholic ♂ ×alcoholic ♀	41	20	4	8	17	12	20	14
Summary	164	64	18	40	82	70	110	82
Control norm. ♂ ×norm. ♀	90	22	2	8	66	19	27	99
♀ treated during pregnancy	4	0	0	0	4	1	1	7
Second generation ×norm.	46	10	3	8, 6 c.e.	33	29, 2 par.	37	25, 3 c.e.
Second gener. ×alcoholic	53	16	8	17, 1 d.e.	29	22, 3 d.e.	39	28
Second gener. ×second gener.	95	29	7	16	59	43, 2 par., 6 d.e.	59	52, 3 d.e., 1 one e., 1 eyeless
Third gener. ×third gener.	48	20	7	14, 1 d. legs	21	19, 1 par., 6 d.e., 2 eyeless	33	13
Third gener. ×second gener.	33	15	4	8	14	16, 1 par., 1 c.e.	24	7
Third gener. ×normal	17	3	4	8	10	5	13	7
Third gener. ×alcoholic	3	1	0	0	2	2	2	1
Second, third gener. ×second, third gener.	18	9	2	6	7	6	12	4

control matings is the fact that from 90 matings only two stillborn litters were produced and, as mentioned above, both of these litters were of so large a size that the mothers seemed unable to successfully deliver them and one of the mothers failed to recover from the process and died a few days later. These two cases make the control records appear worse than they actually should, but in spite of this the control matings have given data equally as good as those generally obtained by careful breeding experiments with vigorous normal stocks. The stock in these experiments is unquestionably good, as the control matings very readily show.

Four normal females were mated and then treated with alcohol throughout their periods of pregnancy and, as the sixth horizontal line of the table indicates, such a treatment was not at all injurious in these particular cases. It actually happened that some of these young were unusually vigorous. The numbers are very small, but this is a direct test, and if such a treatment were really decidedly effective in its action on the embryo or fetus *in utero* these eight young animals should have at least shown some response. It is very possible that after the treatment has been continued for a long time, a year or more, that the mother then presents a uterine environment unfavorable for normal development, since the offspring of such individuals are almost always subnormal. In these cases, however, the inferior quality of the offspring may be due to the action of the alcoholic treatment on the ovarian germ cells rather than the direct environmental effect on the developing embryo or fetus, there is no way at such a stage to separate the two possible effects.

The next three horizontal lines, seventh, eighth and ninth, give the data resulting from the matings in various combinations of the F_1 animals, that is, offspring from alcoholic parentage, but which are not themselves treated with alcohol. The records of these non-treated F_1 individuals are most instructive for an understanding of the actual influences of the alcoholic treatments.

When such F_1 animals are paired with normal individuals the seventh line shows that almost 22 per cent. of the matings failed, which is not a bad record. The proportion of stillborn litters, however, from the F_1 by normal combination was three times as great as from normal matings and 75 per cent. of the stillborn young produced showed gross defects of the eyes, having opaque lenses or typical cataract conditions, while not one of 126 young from normal matings has shown this or any other noticeably abnormal structure. Thirty-three living litters were produced containing in all 54 individuals, 29, or 54 per cent., of which died soon after birth, while 25 survived. Two of those dying soon after birth were paralyzed and unable to walk, while three of the 25 survivors have defective opaque eyes, cases similar to that illustrated by Fig. 1, and many show different nervous symptoms. Thus of 62 full-term young produced by F_1 animals with normal mates, only 25, or 40 per cent., survived for more than a short time after birth, and 12 per cent. of these have gross defects and more than half of them are nervous, excitable individuals, which when mated with normal animals or in any other combination always give very poor quality offspring, if any at all.

The eighth line shows the records of 53 matings between F_1 animals and alcoholics. This combination again gives data comparing most unfavorably with the control and in some ways even worse than the records of matings between two alcoholic animals. Fifteen per cent. of such matings produced stillborn litters! Only one combination gives a worse record of stillborn that is, matings among F_2 animals. Almost half of the young in the living litters died and here again some were deformed. Deformities are strikingly more abundant among the offspring from F_1 and F_2 parents than from the directly alcoholized animals.

The record of 95 *inter se* matings of F_1 animals is shown in the ninth line. Thirty per cent. of such matings gave negative results or early abortions, over 7 per

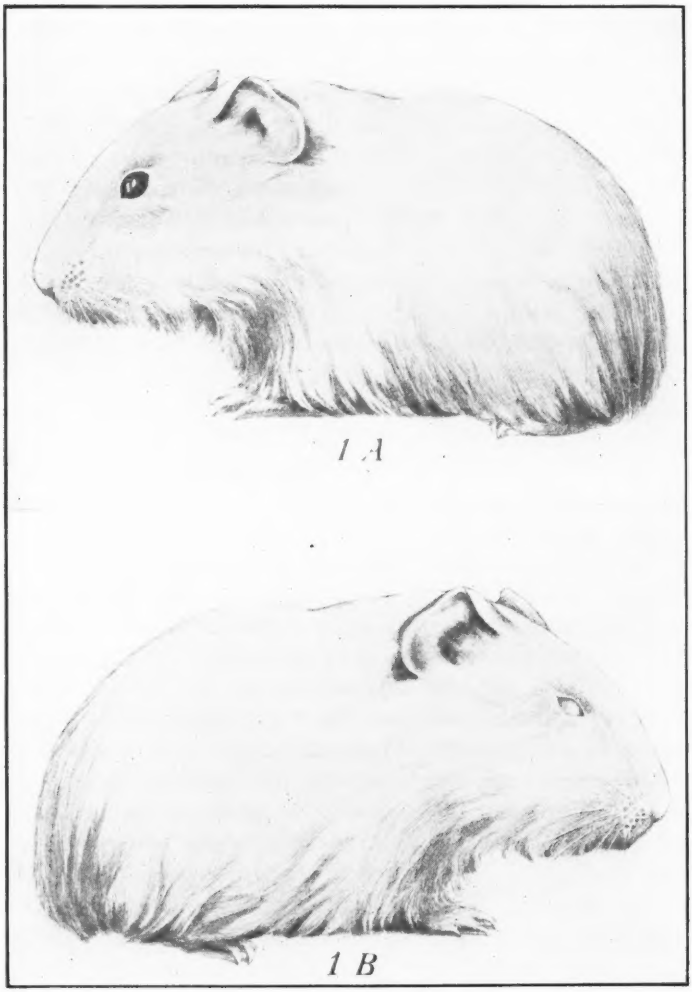

FIG. 1, *A* and *B*. 271 F_2 ♀ (one in litter) (AA)(AN). Both paternal grandparents and the maternal grandfather were alcoholic, no inbreeding. The right eye is smaller than the left and has been entirely opaque since birth. This animal, almost two years old and vigorous, is entirely sterile.

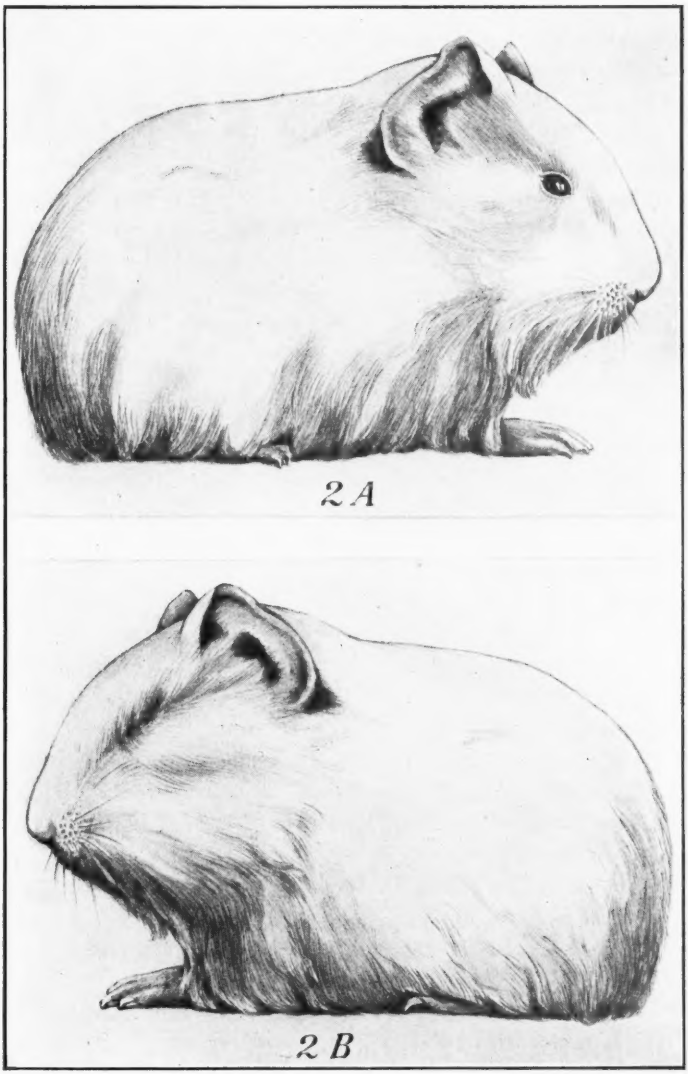

FIG. 2, *A* and *B*. 307 F_2 ♀ (one in litter). Inbred, from brother and sister offspring of an alcoholic male (AN) (AN). The eye of one side normal, the other eye ball apparently absent on living examination. A typical monster monophthalmicum asymmetricum. This animal now 21 months old is completely sterile.

cent. stillborn litters and 62 per cent. living litters. Little less than half of the living young died soon after birth, in all 43, nine of which, or more than one in five, 21 per cent., were paralyzed or deformed; the figures in Plates I and II illustrate the paralytic conditions. Fifty-two of the offspring survived, three with deformed eyes, one with one eyeball completely absent, monster monophthalmicum asymmetricum (Fig. 2, 307 ♀), and almost all of the 52 are very nervous, excitable animals which when bred give rise to deformed or highly degenerate offspring.

The offspring from the F_1 animals mated in any combination are generally far below the normal in power to survive and in quality of structure. When compared with the offspring from directly alcoholized parents, the offspring from the F_1 combinations show an equally bad mortality record and a very much higher proportion of paralyzed and deformed individuals. The 95 matings *inter se* of F_1 animals demonstrate conclusively that such individuals carry defective or abnormal germ cells which give rise to defective developmental products. These degenerate F_2 offspring owe their subnormal condition to the effects of the action of the alcohol treatment upon the germ cells of their grandparents which have been transmitted to them through their parents. In other words, the carriers of hereditary qualities have been modified in the first parental generation, and the effects of this modification are expressed in their offspring F_1, and also in their grandchildren, the F_2 generation.

The next line of the table, the tenth, indicates further how the effects of the original modification are transmitted to the great grandchildren or through three generations since the injury. Forty-eight *inter se* matings of F_2 animals gave the results here shown. Almost 42 per cent. of the matings gave negative results or early abortions, the poorest record in this respect shown in the entire table. About 15 per cent. of such matings gave stillborn litters, 7 in 48 matings, which is remarkably high when compared with any of the above combinations.

The hind legs of one of the stillborn young were deformed in the peculiar manner illustrated in Figs. 4 and 5.

Twenty-one living litters were produced, containing in all 32 young; 19 of these, almost 60 per cent., died soon

FIG. 3. F_3. Two in litter, both same condition, three normal great-grandmothers and three alcoholic great-grandfathers. The parents were single first cousins. Both animals completely eyeless, also with paralysis agitans, one died the second and the other the third day after birth, typical anophthalmia. One brain no indication of optic nerve, the other slight processes.

after birth, and only 13 survived. One of the 19 that died was paralyzed and unable to stand, while 8 of them, a strikingly high proportion, were grossly deformed. Six

had one or both eyes deformed (Figs. 1 and 2), and two were anophthalmic monsters, being completely without eyeballs, optic nerves, optic chiasma or any gross signs of optic tracts (Fig. 3). The brains are now being studied in sections. Figs. 1 to 3 illustrate animals showing the different eye conditions—asymmetrical eyes, monstrum monophthalmicum, and anophthalmic monsters. Figs. 5 and 6 of Plate III illustrate the brains of a normal and an anophthalmic specimen for a comparison of the condition of the optic nerves, etc.

Forty-six full-term young were produced by the F_2 matings, but only 13 of these, or just 28 per cent., were able to survive, while about three times this proportion, or 78.5 per cent. of the full-term young from control matings, survived as vigorous healthy individuals. The 13 living F_3 animals are all rather weak and degenerate and almost completely sterile according to a considerable number of careful matings with strong, fertile guinea pigs. The alcoholic race seems at this stage of the experiment about to fade out in the fourth generation, while normal control lines from the same original stocks have passed far beyond this generation, continuing to breed normally and showing no signs of degeneracy, and never in any case giving rise to a grossly deformed animal.

The eleventh line indicates again the very decided effects transmitted by the descendants of animals which had suffered a modification of their germ plasm by the alcoholization of their tissues. In 33 cases F_1 and F_2 animals were paired together. Fifteen of these matings gave negative results or early abortions, while about 12 per cent. of the matings resulted in stillborn litters of two young each. Only 14 living litters were produced by the 33 matings; these contained in all 23 young, only 7 of which survived. Thus from a total of 31 full-term young only 7, or about 22 per cent., were capable of surviving. All of these young animals are nervous and weak and several offspring from these combinations were deformed.

When F_2 animals are mated with normal individuals

the results are very little if any improved over the two above combinations. Seventeen such matings gave only three failures or early abortions, but a high proportion, 23 per cent., of stillborn litters arose, while 10 living litters, consisting of only 12 individuals, were born. In all 20 full-term young were born and only about one in three of them survived. In this experiment, although one mate was a normal animal, the F_2 mate carried germ cells of so inferior a quality that the output of the combination, admitting the numbers are small, leaves no doubt of the transmission, *through three generations,* of defective conditions induced by alcoholizing the great grandparents of the offspring on only one side of the family, or in only one of the parental lines.

The last line of the table gives the records of mixed combinations of F_1 and F_2 individuals, and here the data are closely similar to those obtained from other combinations of these animals; only about 25 per cent. of the full-term young born are capable of surviving, while 78.5 per cent. of the control young are living.

Briefly, then, 571 matings tabulated in Table I, the records to July 1, 1915, have given rise to 682 full-term young, as well as a large number of premature abortions. A careful study of all these young animals extending over a period of five years has afforded data which convincingly show that the treatment of either the male or the female guinea pig with fumes of alcohol affects the quality of the offspring to which these animals give rise even when paired with normal mates. And further, the changed quality of the offspring is subsequently transmitted through succeeding generations with even more severe marks of degeneration and deformity than those exhibited by the offspring of the directly treated animals.

Other combinations and back crosses are now in progress which are fully in line with the above, but which have not yet afforded sufficient analytical data to record.

The defects caused by the alcohol treatment seem to be largely confined to the central nervous system and organs

of special sense. Paralysis agitans is very common among the F_1, F_2 and F_3 animals. Paralyzed limbs are often observed, the animals being unable to stand or walk (Plates I and II). The eye is also a peculiarly sensitive indicator and presents in the various descendants of alcoholized individuals all degrees of degeneration—

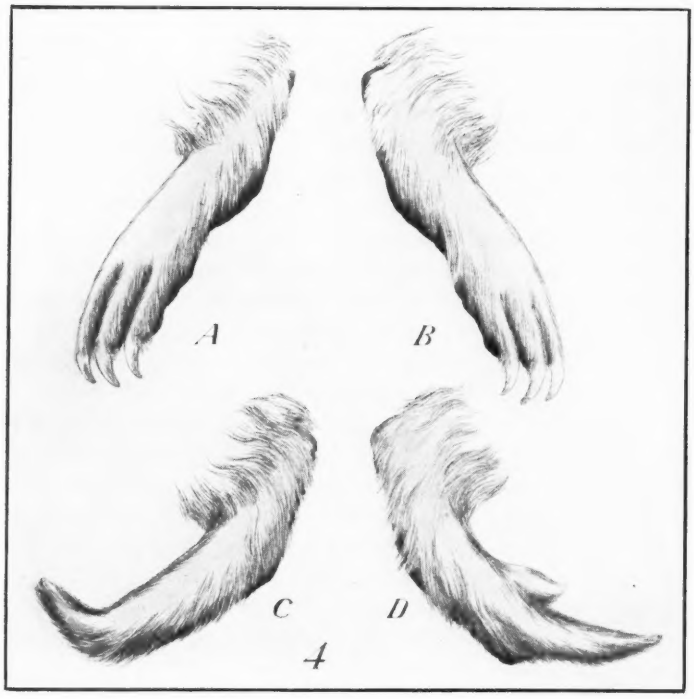

FIG. 4. Hind feet of No. 488 $F_{2,3}$ ♀. All great-grandparents were alcoholic as well as the maternal grandparents. Inbred from mother by son. This animal was one of a litter of two stillborn. The left hind foot, *C*, had only one toe and the right, *D*, one toe and a stump, *A* and *B*, normal right and left hind feet.

opaque cornea, cataract or opaque lenses, small defective eyes, complete absence of one eye and finally complete absence of both eyeballs—anophthalmic monsters. In the latter case the extrinsic eye muscles, the third, fourth and sixth nerves, the lachrymal glands and other struc-

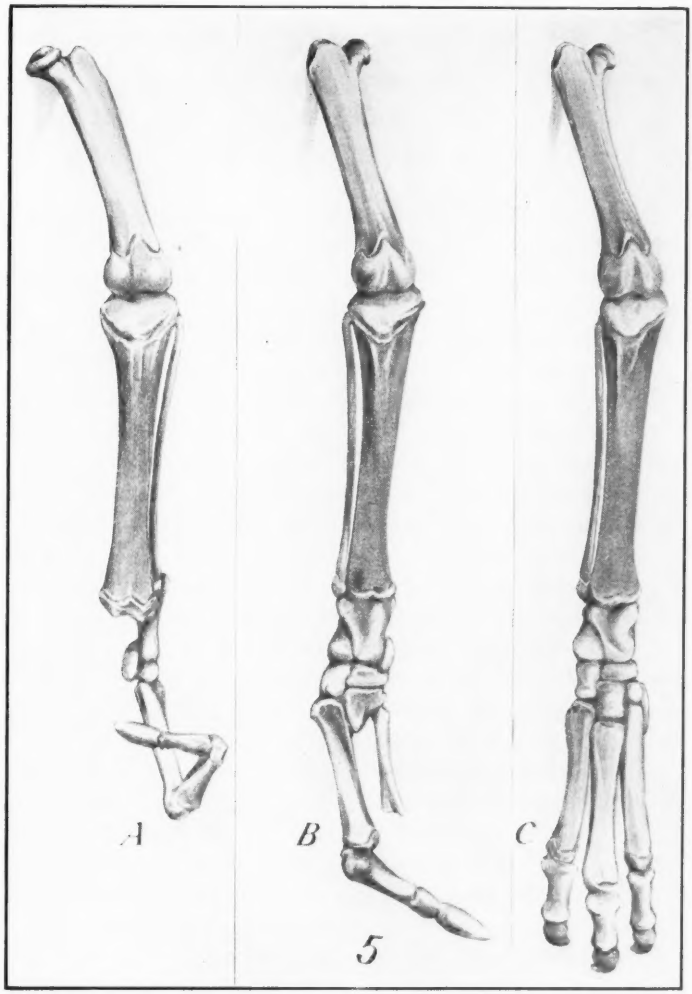

FIG. 5. The skeleton of the two limbs from Fig. 4 shows the left foot, *A*, to have only one metatarsal and toe, the third, and three tarsal bones. The right foot, *B*, has the third toe also and the first metatarsal with the tarsus almost complete. *C* shows the normal skeleton of a right hind limb with the three toes and seven tarsal bones.

tures of the orbit are present, though the eyeball is completely wanting.

Not only are the above congenital eye defects present, but in several instances members of the alcoholic lines have become blind during the first year or year and a half after birth, whereas in our control this has never occurred.

The several illustrations referred to above show specimens exhibiting these various defects. Figs. 1, 2 and 4 of Plates I and II are photographs of animals of indicated lineage which show paralytic conditions. Figs. 4 and 5 illustrate defective extremities. Figs. 1 to 3 show various degrees of defective eyes and absence of eyeball.

It is peculiarly interesting to find these particular eye conditions exhibited by the descendants of alcoholized animals, since, as Stockard ('10) has previously shown, closely similar eye conditions are obtained in great numbers by directly treating the eggs of fish with solutions of alcohol; and like conditions were also obtained, though not so consistently, by treating hens' eggs ('14) with alcohol fumes either before or during incubation.

The table just considered gives only a general idea of the experiment and is in no way analytical. We shall now attempt to analyze these data in such a manner as to determine the influence of internal factors, as, for example, inbreeding on the results. The influence of the size of the litter on the quality of the offspring. The behavior of F_1 and F_2 individuals derived from different lines, and whether there is a difference in the effects on male and female animals, and the manner of transmission of these effects.

(To be continued.)

FECUNDITY IN THE DOMESTIC FOWL AND THE SELECTION PROBLEM[1]

DR. RAYMOND PEARL

I

In the December number of the American Naturalist Professor W. E. Castle[2] directs a vigorous attack against the present writer's work on fecundity. Any one reading Professor Castle's article could scarcely fail, I think, to carry away the impression that the whole of the writer's studies of the past eight years on fecundity in the domestic fowl are to be regarded as essentially valueless. I assume that it was not the intention to convey this impression. The fact, however, appears to be as here stated. With such a conclusion I can scarcely be expected to agree. I shall therefore attempt, in the following pages, in the first place, to call attention to some points regarding my own work which Professor Castle appears to have overlooked, and which seem calculated to give it at least some slight degree of significance, and in the second place, to set forth very briefly my reasons for venturing, in the present state of knowledge, to hold a different opinion from his in regard to some phases of the selection problem.

II

The general plan of Professor Castle's paper appears to be to make a comparison between his selection experiments with rats, and my selection experiments with poultry, to the very great disadvantage of the latter. To this general comparison no general comment on my part can be made, except assent to Castle's conclusion that his

[1] Papers from the Biological Laboratory of the Maine Agricultural Experiment Station, No. 94.

[2] Castle, W. E., "Some Experiments in Mass Selection," Amer. Naturalist, Vol. XLIX, pp. 713–737, 1915.

work on the selection problem is vastly superior to my own. Since the subject of such comparison has been opened it gives me great pleasure to pay tribute, in all sincerity, to Professor Castle's splendid series of experiments on selection in rats. In respect of the numbers of animals involved and their superior adaptability for such an experiment, his work with rats altogether transcends anything which has been done with fowls. These selection experiments constitute an achievement of which their author may well be proud. I have ventured to disagree with Professor Castle's *interpretation* of the results for reasons which will presently be stated. But this difference of opinion, I would most strongly emphasize, concerns *only* the interpretation. We are at one in our high admiration of the factual basis afforded by the rat experiments.

III

Granting all this, however, it seems to me that possibly the case against my studies of fecundity *in toto* is not quite so bad as Castle makes it out to be. Let us examine his points *seriatim*. In the first place the strictures upon the character egg production on p. 714 seem to me to overdo the matter a bit. It is of course true that it is a character confined in its expression to one sex, though that it is also a character which is transmitted by the other sex even Castle somewhat grudgingly admits (p. 715). It also is a character which comes to expression only in the adult. Of this Castle makes a great point throughout his paper, emphasizing that this means that only a small proportion of all offspring born can take part in selection experiments. From the standpoint of methodology this point has nothing like the significance which Castle attributes to it, for the very simple reason that in *all* breeding experiments, his own included, there is a vast amount of random sampling between the population of parental genes and the population of offspring somata. When Professor Castle breeds a pair of rats only a very few

sperm and ova out of the vast hordes the parents produce take part in the production of the resulting litter. He operates, of course, upon the basis that those germ cells which *do* take part in the formation of the litter constitute a *random sample* of the whole population. When I put pullets into the house to test their egg production I operate on precisely the same basis, viz., that I have a random sample of the family from which they are taken. As a matter of fact, I have been at great pains to ensure that the sampling should be random. In all of my studies on the inheritance of fecundity I have regarded this as a point of paramount importance, and have never made use (except occasionally for confirmation of points already made out on other material) of families in which I had not either tested *all* the daughters as to egg production or a sufficiently large random sample to be fairly representative of the family. Further I have repeatedly made careful *ad hoc* investigations of the adequacy and randomness of my sampling.

Castle's next point is, as the matter stands, apparently well taken. He quotes (p. 715) a statement which I somewhat rashly made to the effect "that phænotypic variation of the character fecundity in fowls, markedly transcends, in extent and degree, genotypic variation." Professor Castle's treatment of this statement is perfectly legitimate. If it were true, as stated, it would admit of being turned around as it is in Castle's next sentence, and then it surely would be silly to talk about either selection for this character or about its Mendelian inheritance. What I should have said when I wrote that unfortunate sentence, but did not, was that phænotypic variation *may* transcend genotypic in fecundity, not that it always or regularly does. Because it may I wanted to point out the need for great care in respect of environmental conditions in interpreting results with this character. The real point is this: Long experience in working with winter egg production in poultry has convinced me that under properly controlled environmental conditions this character is as

definitely and regularly controlled by hereditary factors as is the plumage color and pattern. On the other hand, it is a character which is rather particularly sensitive to environmental influences in *one* direction, namely, downward. I can breed a flock of birds which I *know* will be high winter layers if properly fed, housed and managed. But if these birds are starved, housed in a damp cold place and otherwise maltreated they will lay but a few if any eggs. Under such conditions the genotypic condition would be swamped by the environment. It was this sort of thing I had in mind when I made the statement that Castle quotes. It should be particularly noted, however, that this is a somewhat one-sided matter. I can (because I have done so) breed a flock of pullets lacking totally the factors for winter production. With such birds nothing can be done in the way of feeding or management which will make them lay before some time in February or March when the spring cycle begins.

Now all my work on fecundity has been done in a public institution. Egg production is a commercially valuable thing. We have had to submit the results of our breeding operations, in the shape of the birds themselves, to the practical test of farmers, poultrymen, etc. In doing this there has always been vividly before my mind the fact that unless the birds were given proper feed and care, no matter what the genes they carried, they would not lay many eggs.

On the other hand the *degree* of expression of the character in birds carrying the factors for high fecundity may be favorably influenced by exceptionally favorable circumstances, though the possible effects in this direction are, according to my observations, much smaller in amount than in the opposite direction.

While Castle's comments on the unfortunate sentence under discussion are technically perfectly legitimate, I do not think he is quite fair to the essential underlying point of genetic epistemology, namely, the impossibility of judging the genetic constitution by the somatic appearance.

This of course is the reason for the progeny test. I do not think I am in any sense exaggerating if I say that it is one of the chief results of the Mendelian method of studying inheritance to show that in many cases and for many characters it is impossible, in the absence of a progeny test, to be sure of the genetic constitution of the individual from an examination of the soma alone. I fancy that if I cared to be fussily nasty in my controversial methods I could cite page after page from Professor Castle's Mendelian writings where even he, in order to be quite sure about the genetic constitution of an individual, has had to breed it. This is all I mean by the progeny test. Why am I and my fowls held up to scorn and ridicule because I say that it is frequently impossible to tell the genetic constitution of a fowl with respect to fecundity without breeding it? Surely fecundity in poultry and coat color in rats only differ in this respect in degree, *if they differ at all,* not in kind. I think if any one will read pp. 604 and 605 of my last NATURALIST paper, which is the immediate objective of Professor Castle's attack, he will have to admit that the interpretation which I give of the earlier results is not entirely senseless, and might indeed explain them. In any case, it is in thorough accord, methodologically considered, with the very best current Mendelian usage, including that of Professor Castle himself.

IV

This brings us to the most serious phase of Castle's attack, namely that in which he denies the validity of my conclusions respecting the inheritance of the character fecundity in fowls.

On the top of p. 716 he asserts that I "assume" that two Mendelizing factors are concerned in the inheritance of fecundity, "but without any sufficient published evidence for either conclusion." As I have published[3] many pages of evidence in demonstration of my conclusions on this point, one can only infer from this statement of

[3] In particular in the *Journal of Experimental Zoology,* Vol. 13, 1912.

Castle's that he regards that evidence as totally worthless. It has not so appealed to other workers.[4] Furthermore I think it can be shown that methodologically my treatment of the problem of inheritance of fecundity stands on precisely the same plane as Mendelian work in general, and Professor Castle's Mendelian work in particular. This I shall now try to do.

The essence of a test of a Mendelian hypothesis lies in this: the genetic constitution of the parents of an array of offspring necessitates that the individual offspring bearing different segregating characters, or different segregating categories of the same character, shall occur in definite numerical proportions. If the observed numerical proportions of the offspring agree, within the limits of error due to random sampling, with the proportions expected from the Mendelian hypothesis, then this fact constitutes valid evidence in support of the hypothesis. If no exceptions to this rule appear and a sufficient number of agreeing cases are adduced the hypothesis is regarded as demonstrated. The number of cases necessary to constitute a proof is a purely individual matter. What one person will consider sufficient to establish proof another will not.

Now in the case of fecundity in fowls, Pearl and Surface[5] first established that the Barred Plymouth Rock stock at the Maine Experiment Station was not homozygous in respect of winter egg production, but that it contained, with frequent occurrence, individuals of high fecundity, and also individuals of low fecundity. The *race* not being homozygous with respect to fecundity, it was possible to test the Mendelian inheritance of this

[4] Cf., for example, Morgan, T. H., "Heredity and Sex," New York, 1913, Doncaster, L., "The Determination of Sex," Cambridge, 1914, and Johannsen, W., "Elemente der exakten Erblichkeitslehre," Zweite Ausgabe, Jena, 1913, Plate L, Vererbungslehre, Leipzig, 1913, Brown, E., "Poultry Husbandry," London, 1915, Sturges, T. W., "The Poultry Manual," 3d edition, London 1915.

[5] Pearl and Surface, "Data on the Inheritance of Fecundity Obtained from the Records of Egg Production of the Daughters of '200-Egg' Hens," Me. Agr. Exp. Sta. Bull. 166, 1909.

character *within* the race, without crossing, by the above scheme.

The next step was the definition of the categories of the character winter egg production. From long study of the character I concluded that the natural categories in this strain were (*a*) zero winter production, (*b*) winter production between zero and 30 eggs, and (*c*) a winter production of over 30 eggs. These were chosen as working categories. If any one will turn to p. 719 of Professor Castle's paper and examine Fig. 1, which is there printed, they will find that even he chooses categories of the character with which he is working. Nowhere have these ever been quantitatively defined; nowhere has he ever presented any evidence that the step from his rat grade + 1 (for example) to his grade + 2 represents a more or less inclusive category than a difference in winter production of from 0 to 30 eggs. Professor Castle reads us a beautiful little homily about Mendel's peas. But I am not clear that either Mendel or Castle has shown that the amount of variation *within* the category "yellow" is less than the amount of variation within my fecundity category of "under 30." From the only study which has ever been made of the matter, Weldon's,[6] I should certainly conclude that the category "under 30" in winter egg production carries within itself distinctly *less* variation than the category "yellowness" in peas. Castle's assertion about my fecundity categories ill becomes one whose work in genetics has dealt almost without a single exception with non-quantitatively defined Mendelian categories. Of course, as a matter of fact, he knows, I know, and everybody knows that the variations within the Mendelizing category are of no significance so far as the Mendelian result is concerned. I happen to have observed, for example, that there are at least four genetically distinct rose combs in poultry. Yet they are all *rose;* any of them crossed with single gives a 3:1 ratio in F_2.

[6] Weldon, W. F. R., "Mendel's Laws of Alternative Inheritance in Peas," *Biometrika,* Vol. I, pp. 228–265, 1902.

Having chosen these categories of the character fecundity because they appeared to represent natural divisions, I proceeded to show for hundreds of matings the distribution of the progeny when individual females whose performance fell into one or another of the categories were mated to particular males. This was done both for the pure bred Barred Rocks and for crosses. The results at once showed that definite ratios were appearing with regularity and constancy. Further analysis showed that a Mendelian hypothesis which postulated two factors, one sex-linked and the other not, accounted for all the facts.

If all this does not conform to the classic canons of Mendelian experimentation, I am sure I do not know what does.

V

Castle charges me with suppressing data. There are just two things which I wish to say regarding this charge. The first is that I shall publish the complete raw data of my work on the inheritance of fecundity when I have finished my own study of these, and not sooner. I am using this material for the study of various problems. There appears to be no reason why I should make valuable original records public property until such time as I have finished my own analysis of them. If Professor Castle will examine my published papers he will find that in lines of work which I am finishing and leaving, complete raw data are published (cf. for example "A Biometrical Study of Egg Production in the Domestic Fowl," Parts I to III).

In the second place I wish to say that so far as any question of concealment is concerned Professor Castle, or any of his students, will be very welcome to come to the laboratory at any time, for as long as they like, and make any examination of the original record books in connection with published results and conclusions.

There is one further point which needs consideration concerning the charge of suppression of pertinent facts. An important reason, I think, why Professor Castle's own interpretation of his rat selection experiment has not been

freely and universally accepted by workers in genetics lies in the fact that he has never presented his results in such a form that any other interpretation of the data could by any chance be tested. There is, from the methodological standpoint, only one way in which an adequate test can be made as to whether any observed change in the composition of a population is the result of a sorting, or of true germinal change, or an adequate idea gained of *how* the change came about. This is the method of individual pedigree analysis. Only one extensive mass selection experiment has ever been analyzed in this way, and that is in Surface's[7] discussion of the Illinois corn results. The Hagedoorns[8] called Castle's attention two years ago to the necessity of individual pedigrees before any just opinion could be formed as to the meaning of the data. To paraphase Castle's damning indictment of the present writer I may be permitted to call attention to the fact that, so far as concerns the individual pedigree of his rats, "information is denied us" by Castle.

In bringing to a close this part of the discussion I wish to emphasize that, in spite of Castle's assertion to the contrary, any unprejudiced person who will take the trouble to examine the facts will find that, so far as concerns methods of dealing with the data and presenting them for publication, the method of their Mendelian analysis, the method of presenting the results of selection experiments by a series of averages, and other matters of method, my work with fecundity in fowls exactly parallels at every point Castle's work with hooded rats, and is in every way, so far as I am able to judge, exactly as critical as his. His experiments are more extensive in scope than mine, and the character fecundity is a more difficult one to deal with, but so far as methodology is concerned the two researches stand on precisely the same footing. I have not

[7] Surface, F. M., "The Result of Selecting Fluctuating Variations." Data from the Illinois Corn Breeding Experiments. IV[e] Conf. int. de Gen., pp. 221–256, 1911.

[8] Hagedoorn, A. L. and A. C., "Studies on Variation and Selection," *Zeitschr. f. ind. Abst.—und Vererbungslehre*, Bd. XI. pp. 145–83, 1914.

lumped the data any more, nor have I "suppressed" data any more than he has. On the contrary I have published a great deal of exact data, in a series of papers from this laboratory, regarding the character fecundity, its normal variation, etc.

VI

The next point which Castle makes is that the changes which occurred in mean flock production during the sixteen years, for which figures were given in the paper which he criticizes, were probably due to environmental, or at least to non-genetic effects. In making this point he calmly disregards all that I have ever published about the experiments, the means taken to be sure that environmental effects were not mistaken for genetic, etc., and proceeds in his discussion as though all my work on the subject had been absolutely uncritical and that I had never given a thought to checking the correctness of the results. In the first place he notes the changes in the numbers of birds on which the average in different years are based, and points out that these numbers change in a roughly inverse direction to the means. He then says:

> Has not the better environment and lessened competition of small numbers possibly something to do with the result?

They have not. Had Professor Castle been less eager to demolish these fecundity results he might have noted that I have repeatedly stated that since 1908 *all birds in these experiments have been kept in flocks of the same size,* namely 125 birds per flock. The number of such flocks has at times varied, but not the number in each flock[9] except by very small numbers, such as resulted from losses by death, the necessity occasionally of putting a few extra birds in a pen for a brief period and similar very minor

[9] To prevent any mental strain in reconciling the above statement with the third column of Table I, p. 599, in my NATURALIST paper, let me hasten to say that the pens were filled out, if the number of Barred Rocks in the selection experiments did not just equal multiples of 125, with birds from other experiments.

fluctuations. In the first four years (1899–1900, 1900–1901, 1901–1902, 1902–1903) of the *old* experiment the birds were kept in 50-bird flocks. During the five years following (*i. e.*, to 1908–1909) they were kept in 50, 100, and 150 bird flocks. Just precisely how much (or really how little) difference the size of flock made in average egg production has been fully and minutely analyzed biometrically and published by Pearl and Surface[10] some six years ago. It seems reasonable to suggest that before indulging in fast and loose criticism on such a simple point of fact as this it would become Professor Castle to read the literature respecting the work he is attacking. Since this material seems to have been forgotten it may be well to repeat here that the results showed (Pearl and Surface *loc. cit,* p. 115) that in general there was no significant difference in *winter* production between 50, 100, and 150 bird flocks. In later months of the laying year differences appeared but only in the last month of the winter period (February) was there any significant excess of even 50-bird flocks over the others. Furthermore, besides the material which has already been published regarding the possible influence of environmental factors on the results of these experiments, I have carried out a number of special investigations on different phases of this general question which have not yet been published. For example, I have minutely analyzed the data regarding date of hatching to see whether that might not enter as a significant factor in the interpretation of the results. The data on this question are being prepared for publication now, but it may be said in advance that the results show that date of hatching can not possibly have had anything to do with the rise in average flock production which has occurred between 1908 and 1915.

[10] Pearl and Surface, "A Biometrical Study of Egg Production in the Domestic Fowl. Part I. Variation in Annual Production. U. S. Dept. of Agr., Bu. A. I. Bull. 110, pp. 1–80, 1909. Also the effect of flock size upon *winter* production is specifically discussed in detail in Part II, of the same "Study," pp. 113–117, 1911.

VII

Turning now to the general problem of selection there are certain fundamental matters which it seems to me are in danger of being lost sight of in the rapid shiftings of view point which are an essential part of any general controversial campaign, such as Professor Castle's writings of the last few years would indicate that he engaged in. These are:

1. The pure-line concept has certainly been one of the most useful working tools in the practical breeding of plants and animals that has ever appeared. Particularly in plant breeding the pioneer work at Svalöf, which has been repeated and duplicated on a most extensive scale in plant breeding laboratories all over the world, demonstrates in the most complete manner that, whatever may be happening in the germ-plasm of rats, certainly the germ-plasm of our common cereal crops is in such a state or condition that selection within the pure line is without effect. This is a *fact,* real and definite. It lies definitely at the basis of very extensive commercial seed breeding operations in various different countries. To any one familiar with the extent and stability of the practical applications of the pure-line concept in cereal breeding operations, some of our current discussions of the selection problem seem very academic indeed. Even the justly celebrated magnitude of Castle's rat experiments is scarcely of the same order as the combined and accordant experience of expert cereal breeders throughout the world. Before any one makes up his mind finally about the problem of the efficiency of selection within the pure line it will be well to remember that besides Johannsen's famous, if now in certain quarters somewhat distrusted, beans, there are all the Svalöf oats, wheats, etc., to be reckoned with.

2. No one has ever disputed the power of systematic selection to alter populations, which were not pure-lines. Such alteration may extend the *range* of variation very greatly beyond what it was in the original population.

From a methodological standpoint, however, it is necessary to have a very different sort of evidence from that afforded by changing general population means, such as Castle gives for his rats, and I for fecundity, to prove that the process of selection has been the cause of a change in the absolute somatic equivalent of a particular gene or hereditary determinant.

3. It is just in connection with this last point that there seems to me to be a good deal of unclear thinking and arguing at cross-purposes about the selection problem. Let us examine the logic of the matter symbolically.

Let there be a character A, whose somatic variation in the general population is given by a frequency distribution of area $Z \backsimeq \frac{A_n}{A_1} A$, where Z is the frequency of occurrence of the somatic state or condition A_1, and so on to Z_n and A_n. Now suppose that selection is practised for the somatic condition A_{40}, but that in the original population A_{38} is the most extreme variation in that direction found to exist. Then for A_{40}, $Z_{40}=0$, and for A_{38}, Z_{38} is very small. Let it be further supposed that the somatic difference between the A_{38} and A_{39} condition may be of *any* determinate magnitude R. It makes no difference to the logic of the case whether R is large or is extremely minute. Now suppose, as a limiting case, that we assume a gamete-soma correlation of 1, *i. e.*, perfect. Then in the gonads of an individual somatically A_{38}, all the germ cells will bear the factor a_{38}. If two such individuals are bred together the progeny will be somatically A_{38}.[11] Suppose that for m generations the matings are of $A_{38} \times A_{38}$. This is continued selection. Then suppose in the $m+1$th generation, *ex* $A_{38} \times A_{38}$ parents, appears an A_{39} individual.

Concretely this represents a step in advance in the direction of selection. Let us analyze the possible ways in which this may have happened.

[11] This is precisely the condition which prevails in a pure line of oats, except for purely phænotypic variation, superimposed by environmental factors.

(*a*) First we may assume that A_{38_m} and A_{38_m}, the parents of this $A_{39_{m+1}}$, instead of having a_{38} gametes had a_{39} gametes. This would correspond to what is called a mutation. The gamete-soma correlation has been broken by the appearance of a new kind of gamete different from the parental gametes. There has been a sudden definite change in the germ plasm, such that an a_{38} germ plasm has changed to an a_{39} germ plasm.

(*b*) Or, we may assume that $A_{39_{m+1}}$ was produced by the union of two a_{38} gametes, but that these gametes dedevelop a 39 soma instead of a 38 soma. This assumption leads logically straight to genetic indeterminism, a conclusion which, I think, is repugnant to all that is known regarding the physiology of the hereditary process.

Embracing alternative (*a*) then, we may next inquire as to the possible cause of this sudden change of the germ plasm, by an amount of which the somatic equivalent is R, from a_{38} to a_{39}. If we say that this change has been *caused* by a selection, we can only conclude that the fact that A_{38_1}, A_{38_2} . . . A_{38_m} have been placed in particular cages or apartments to breed, for this is the only physical thing that selection means in this case, has been the *cause* of the germinal change. For by hypothesis there has been no mixing of germ-plasms. We have been practising straight selection of the most extreme somatic individuals, all by hypothesis A_{38}, and each homozygous. It seems to me a misuse of terms to say in such a case as that postulated that *selection* has caused the appearance of the *variation* which it selects, unless we are prepared to say that the physical act of the selection of the individuals for mating physiologically effects the germ plasm. Such an assumption we are all agreed would be nonsense. What has happened in the postulated case is precisely this: a new heritable variation in the direction of selection *has appeared* while selection was in progress. If we say any more than this we are going beyond our facts. If the selectionist would state his results in this form, and

not incessantly harp on the string that "selection *caused*" his results, he would be on logically solid ground and would receive a more respectful hearing from those who place a high value upon clear thinking and sound logic in scientific matters.

Now up to this point in the argument there has been no biological point involved, so far as I can see, to which anybody, whether of the pure line or the selection faith can take exception. Certainly I am perfectly willing to admit that germ-plasm changes do sometimes occur, of all magnitudes from the most minute up. Further no one, I take it, will deny that, having appeared, these variations may be seized upon and preserved by selection. I do desire to emphasize, however, that there is no evidence, as yet, that the selection *causes* the variations.

It may be objected that the postulated case is too simple and leaves out of account too many factors. All this, however, will not affect the logic of the case. Generalized, that logic is as follows: A heritable difference between two individuals or races implies a difference in the germ plasm. The difference in the germ plasm must have made its initial appearance at a definite point of time. At that time the germ plasm *changed* from its previous condition. The cause of that change can not be conceived to be the selection for breeding purposes of the parents bearing the unchanged germ plasm. To assert that the new variation is a result of amphimixis due to mating unlike parents would be, in the present state of genetic knowledge, a ridiculous begging of the question, because, in the first place, by hypothesis in any selection experiment individuals genetically as nearly alike as possible are always mated together, and in the second place, as selection continues homozygosity automatically increases.

The whole fact of the matter is that the assertion that selection *per se* causes changes in the germ plasm, is a wholly new addition to the classic Darwinian selection theory, tacked on quite inadvertently, I believe, by some of the modern exponents of that theory. Darwin never

supposed that selection was a cause of favorable variation. Instead he repeatedly pointed out that the fundamental problem behind natural selection was that of the cause of the variations which selection preserved. That problem remains to-day practically in the same condition that it was left by Darwin. We are no nearer, essentially, now than we were then to knowing the cause of *new* variations. The assertion that new variations are caused by selection is the rankest kind of mysticism plus bad logic.

But if selection of the parents can not be supposed the cause of new variations in the individual, then clearly what selection does, and all it can do, is to change the germinal constitution of a race or population by preserving those individuals in which new variations have appeared, and multiplying them. This is exactly what has been done in the hooded rat experiment, it seems to me on Castle's interpretation of the case. In that experiment every favorable variation in the many thousands of rats has been preserved and the individuals bearing it have been multiplied. Others have been thrown away. The range of the character in the direction of selection has been extended far beyond the original range. But would it have been so extended, or could it have been, if the favorable variations had not appeared for selection, or if, having appeared, they had not been heritable? Suppose one started such an experiment with a character which was in a stable condition and not varying. Take, for example, the single comb of fowls, and attempt by selection from a pure single-combed race to produce a stable rose-combed race by selection alone. Prophecy is dangerous business, but I do fancy one would be a very long time on that job! Characters, so far as I can see, will be altered following selection just in proportion as they are varying genotypically. The cause of the *alteration* is to be sought in the cause of the *variations*, not in the selection only.

I have for some time felt that probably the differences in opinion between the selectionists, as represented by

Castle, and the advocates of the pure-line concept, reduces itself finally very largely to a dispute over the use of words, if both are discussing the same objective facts or experiments. It is repugnant to the logical faculties of the pure-linists to be told that selection is a cause of new variations. On the other hand, I suspect that this particular use of words, which is offensive to our camp, would not be deemed absolutely essential to the making of their case by Castle and his followers. Castle's special *bête noir* appears to be that the pure-linists seem to him to deny the possibility of germinal variation, except it be large in amount (a proper De Vriesian mutation). Now I am in no wise authorized to speak for the pure-line advocates, but I can say for myself, and I venture to think others would agree, that this contention forms no part of the real, genuine pure-line body of doctrine. The followers of the pure-line merely have observed *in fact* that it is not so easy to change all things by a process of selective breeding as it has been to change the pattern of Castle's rats, or the egg production of my fowls. Many characters, and many organisms, when got into a homozygous condition exhibit any germinal variation so rarely as to make any change by the selection of such variation impossible within the limits of finite experimentation. Neither Johannsen nor any followers of his, so far as I am aware, have ever attempted to set any limitations on how big or how little a germinal variation could be.

THE EVOLUTION OF THE CELL. II

By the Late PROFESSOR E. A. MINCHIN, F.R.S.

Even more remarkable than the relation of the chromosomes to cell-reproduction is their behavior in relation to sexual phenomena. In the life-cycles of Metazoa the sexual act consists of the fusion of male and female pronuclei, each containing a definite and specific number of chromosomes, the same number usually, though not always, in each pronucleus. It has been established in many cases, and it is perhaps universally true, that in the act of fertilization the male and female chromosomes remain perfectly distinct and separate in the synkaryon or nucleus formed by the union of the two pronuclei, and, moreover, that they continue to maintain and to propagate their distinct individuality in every subsequent cell-generation of the multicellular organism produced as a result of the sexual act. In this way, every cell of the body contains in its nucleus distinct chromatinic elements which are derived from both male and female parents and which maintain unimpaired their distinct and specific individuality through the entire life-cycle. This distinctness is apparent at least in the germ-cell-cycle of the organism, but may be obscured by secondary changes in the nuclei of the specialized tissue-cells.

Only in the very last stage of the life-cycle do the group of male and female chromosomes modify their behavior in a most striking manner. In the final generation of oogonia or spermatogonia, from which arise the oocytes and spermatocytes which in their turn produce the gamete-cells, it is observed that the male and female chromosomes make a last appearance in their full number, and then fuse in pairs, so as to reduce the number of chromosomes to half that previously present.

In *Aggregata* also Dobell and Jameson have shown that the union of the pronuclei in fertilization brings together two sets each of six chromosomes, and that these then fuse with one another in pairs according to type, that is to say *a* with *a*, *b* with *b*, *c* with *c*, and so on. Analogous phenomena have been demonstrated also in the gregarine *Diplospora*. We have here a difference in detail, as compared with the Metazoa, in that the fusion takes place at the fertilization and not as the first step in the maturation of the germ-cells; but in both cases alike the fusion of chromatin-elements individually distinct and exhibiting specific characteristics is to be regarded as the final consummation of the sexual act, though long deferred in the Metazoan life-cycle.

As Vejdovský has pointed out, there can be no more striking evidence of the specific individuality of the chromosomes than their fusion or copulation in relation to the sexual act. Is there any other constant element or constituent of living organisms exhibiting to anything like the same degree the essentially vital characteristics of individuality manifested in specific behavior? If there is, it remains to be discovered.

I come now to the question of the permanence and immortality, in the biological sense of the word, of the chromatinic particles, which may be summarily stated as follows: the chromatinic particles are the only constituents of the cell which maintain persistently and uninterruptedly their existence throughout the whole life-cycle of living organisms universally.

I hope I shall not be misunderstood when I enunciate this apparently sweeping and breathless generalization. I am perfectly aware that in the life-cycle of any given species of organism there may be many cell-constituents besides the chromatin-particles that are propagated continuously through the whole life-cycle; but cell-elements which appear as constant parts of the organization of the cell throughout the life-cycle in one type of organism may be wanting altogether in other types. With the exception

of the chromatin-particles there is no cell-constituent that can be claimed to persist throughout the life-cycles of organisms universally. To take some concrete examples; the cytoplasmic grains known as mitochondria or chrondriosomes have been asserted to be persistent elements throughout the germ-cycle of Metazoa, and the function of being the bearers of hereditary tendencies has been ascribed to them. But Vejdovský[22] flatly denies the alleged continuity in cases investigated by him, and though chrondriosomes have been described in some Protozoa, there is no evidence whatever that they are of universal occurrence in Protista. Centrosomes, intranuclear or extranuclear, have been stated to be constant cell-components in some organisms; whether that is true or not it seems quite certain that in many organisms the cells are entirely without centrosomic bodies of any kind, as for example in the whole group of Phanerogams. So it is with any other cell-constituent that can be named. It may be that this is only the result of our incomplete knowledge at the present time. I am prepared, however, to challenge anyone to name or to discover any cell-constituent, other than the chromatinic particles, which are present throughout the life-cycle, not merely of some particular organism, but of organisms universally.

In this feature of continuity the chromatin-constituents of the cell present a remarkable analogy with the germplasm of Metazoa. Just as the germ-cells of Metazoa go on in an uninterrupted, potentially everlasting series of cell-generations, throwing off, as it were, at each sexual crisis a soma which is doomed to but a limited lease of life, during which it furnishes a nutritive environment for further generations of germ-cells; so in the series of cell-generations themselves, whether in the germ-cell-cycles of Metazoa or in the life-cycles of Protista the chromatin-particles maintain an uninterrupted propagative series within a cell-body of which the various parts have a limited duration of existence, making their appearance, flourish-

[22] *L. c.*, pp. 77–89.

ing for a time, and disappearing again. This analogy between the chromatin of cells and the germ-plasm of multicellular organisms becomes still more marked when we find that in many Protozoa the chromatin may undergo a specialization into generative and trophic chromatin, the former destined to persist from one life-cycle to another, the latter destined to control cell-activities merely during one cycle, without persisting into the next. The differentiation of generative and trophic chromatin is now well known to occur in many Protozoa, and in its most extreme form, as seen in the Infusoria, it is expressed in occurrence of two distinct nuclei in the cell-body.

To recapitulate my argument in the briefest form; the chromatinic constituents of the cell contrast with all the other constituents in at least three points: physiological predominance, especially in constructive metabolism; specific individualization; and permanence in the sense of potential biological immortality. Any of these three points, taken by itself, is sufficient to confer a peculiar distinction to say the least, on the chromatin-bodies; but taken in combination they appear to me to furnish overwhelming evidence for regarding the chromatin-elements as the primary and essential constituents of living organisms, and as representing that part of a living body of any kind which can be followed by the imagination, in the reverse direction of the propagative series, back to the very starting-point of the evolution of living beings.

In the attempt to form an idea as to what the earliest type of living being was like, in the first place, and as to how the earliest steps in its evolution and differentiation came about, in the second place, we have to exercise the constructive faculty of the imagination guided by such few data as we possess. It is not to be expected, therefore, that agreement can be hoped for in such speculations; it would indeed be very undesirable, in the interests of science, that there should be no conflict of opinion in theories which, by their very nature, are beyond any possibility of direct verification at the present time. The

views put forward by any man do but represent the visions conjured up by his imagination, based upon the slender foundation of his personal knowledge, more or less limited, or intuition, more or less fallacious, of an infinite world of natural phenomena. Consequently such views may be expected to diverge as widely as do temperaments. If, therefore, I venture upon such speculations, I do so with a sense of personal responsibility and as one wishing to stimulate discussion rather than to lay down dogma.

To me, therefore, the train of argument that I have set forth with regard to the nature of the chromatinic constituents of living organisms appears to lead to the conclusion that the earliest living beings were minute, possibly ultramicroscopic particles which were of the nature of chromatin. How far the application of the term chromatin to the hypothetical primordial form of life is justified from the point of view of substance, that is to say in a biochemical sense, must be left uncertain. In using the term chromatin I must be understood to do so in a strictly biological sense, meaning thereby that these earliest living things were biological units or individuals which were the ancestors, in a continuous propagative series, of the chromatinic grains and particles known to us at the present day as universally-occurring constituents of living organisms. Such a conception postulates no fixity of chemical nature; on the contrary, it implies that as substance the primitive chromatin was highly inconstant, infinitely variable, and capable of specific differentiation in many divergent directions.

For these hypothetical primitive organisms we may use Mereschkowsky's term biococci. They must have been free-living organisms capable of building up their living bodies by synthesis of simple chemical compounds. We have as yet no evidence of the existence of biococci at the present time as free-living organisms; the nearest approach to any such type of living being seems to be furnished by the organisms known collectively as Chlamydozoa, which up to the present have been found to occur

only as pathogenic parasites. In view, however, of the minuteness and invisibility of these organisms, it is clear that they could attract attention only by the effects they produce in their environment. Consequently the human mind is most likely to become aware in the first instance of those forms which are the cause of disturbance in the human body. If free-living forms of biococci exist, as is very possible and even probable, it is evident that very delicate and accurate methods of investigation would be required to detect their presence.

I am well aware that the nature and even the existence of the so-called Chlamydozoa is uncertain at the present time, and I desire to exercise great caution in basing any arguments upon them. In the descriptions given of them, however, there are some points which, if correctly stated, seem to me of great importance. They are alleged to appear as minute dots, on the borderline of microscopic visibility or beyond it; they are capable of growth, so that a given species may be larger or smaller at different times; their bodies stain with the ordinary chromatin-stains; and they are stated to reproduce themselves by a process of binary fission in which the body becomes dumbbell-shaped, appearing as two dots connected by a slender thread, which is drawn out until it snaps across and then the broken halves of the thread are retracted into the daughter-bodies. This mode of division, strongly reminiscent of that seen in centrioles, appears to me to permit of certain important conclusions with regard to the nature of these bodies; namely, that the minute dot of substance has no firm limiting membrane on the surface and that it is of a viscid or semi-fluid consistence.

If it be permissible to draw conclusions with regard to the nature of the hypothetical biococci from the somewhat dubious, but concrete data funished by the Chlamydozoa, the following tentative statements may be postulated concerning them. They were (or are) minute organisms, each a speck or globule of a substance similar in its reactions to chromatin. Their substance could be described

as homogeneous with greater approach to accuracy than in the case of any other living organism, but it is clear that no living body that is carrying on constructive and destructive metabolism could remain for a moment perfectly homogeneous or constant in chemical composition. Their bodies were not limited by a rigid envelope or capsule. Reproduction was affected by binary fission, the body dividing into two with a dumbbell-shaped figure. Their mode of life was vegetative, that is to say, they reacted upon their environmental medium by means of ferments secreted by their own body-substance. The earliest forms must have possessed the power of building up their protein-molecules from the simplest inorganic compounds; but different types of biococci, characterized each by specific reactions and idiosyncrasies, must have become differentiated very rapidly in the process of evolution and adaptation to divergent conditions of life.

Consideration of the existing types and forms of living organisms shows that from the primitive biococcal type the evolution of living things must have diverged in at least two principal directions. Two new types of organisms arose, one of which continued to specialize further in the vegetative mode of life, in all its innumerable variations, characteristic of the biococci, while the other type developed an entirely new habit of life, namely a predatory existence. I will consider these two types separately.

(1) In the vegetative type the first step was that the body became surrounded by a rigid envelope. Thus came into existence the bacterial type of organism, the simplest form of which would be a Micrococcus, a minute globule of chromatin surrounded by a firm envelope. From this familiar type an infinity of forms arises by processes of divergent evolution and adaptation. With increase in size of the body the number of chromatin-grains within the envelope increase in number, and are then seen to be imbedded in a ground-substance which is similar to cytoplasm, apparently, and may contain non-chromatinic en-

closures. With still further increase of size the chromatin-grains also increase in number and may take on various types of arrangement in clumps, spherical masses, rodlets, filaments straight or twisted in various ways, or even irregular strands and networks,[23] and the cytoplasmic matrix, if it is correct to call it so, becomes correspondingly increased in quantity. I will not attempt, however, to follow up the evolution of the bacterial type further, nor to discuss what other types of living organisms may be affiliated with it, as I have no claims to an expert knowledge of these organisms. I prefer to leave to competent bacteriologists and botanists the problem of the relationships and phylogeny of the Cyanophyceæ, Spirochætes, etc., which have been regarded as having affinities with Bacteria.

(2) In the evolution from the biococcus of the predatory type of organism, the data at our disposal appear to me to indicate very clearly the nature of the changes that took place, as well as the final result of these changes, but leave us in the dark with regard to some of the actual details of the process. The chief event was the formation, round the biococci of an enveloping matrix of protoplasm for which the term periplasm (Lankester) is most suitable. The periplasm was an extension of the living substance which was distinct in its constitution and properties from the original chromatinic substance of the biococcus. The newly-formed matrix was probably from the first a semi-fluid substance of alveolar structure and possessed two important capabilities as the result of its physical structure; it could perform streaming movements of various kinds, more especially amœboid movement; and it was able to form vacuoles internally. The final result

[23] See especially Dobell, "Contributions to the Cytology of the Bacteria," *Quart. Journ. Micr. Science*, LVI (1911), pp. 461, 462. I can not follow Dobell in applying the term "nuclei" to these various arrangements of the chromatin-grains in Bacteria. Vejdovsky compares them with chromosomes; but there is no evidence that they play the part in the division and distribution of the chromatin-grains which is the special function of chromosomes, as will be discussed in more detail presently.

of these changes was a new type of organism which, compared with the original biococci, was of considerable size, and consisted of a droplet of alveolar, amœboid periplasm in which were imbedded a number of biococci. Whether this periplasm made its first appearance around single individual biococci, or whether it was from the first associated with the formation of zooglœa-like colonies of biococci, must be left an open question.

Thus arose in the beginning the brand of Cain, the prototype of the animal, that is to say, a class of organism, which was no longer able to build up its substance from inorganic materials in the former peaceful manner, but which nourished itself by capturing, devouring, and digesting other living organisms. The streaming movements of the periplasm enabled it to flow round and engulf other creatures; the vacuole-formation in the periplasm enabled it to digest and absorb the substance of its prey by the help of ferments secreted by the biococci. By means of these ferments the ingested organisms were killed and utilized as food, their substance being first broken down into simpler chemical constituents and then built up again into the protein-substances composing the body of the captor.

A stage of evolution is now reached which I propose to call the pseudo-moneral or cytodal stage, since the place of these organisms in the general evolution of life corresponds very nearly to Haeckel's conception of the Monera as a stage in the evolution of organisms, though not at all to his notions with regard to their composition and structure. The bodies of these organisms did not consist of a homogeneous albuminous "plasson," but of a periplasm corresponding to the cytoplasm of the cell, containing a number of biococci or chromatin-grains. Thus their composition corresponded more clearly to that of plasson as conceived by Van Beneden, when he wrote: 'Si un noyau vient à disparaître dans une cellule, si la cellule redevient un cytode, les éléments chimiques du noyau et du nucléole s'étant repandus dans le protoplasme, le plasson se trouve

de nouveau constitué.' If we delete from this sentence the word "chimiques" and also the words "et du nucléole," and substitute for the notion of the chemical solution of the chromatin-substance that of scattered chromatin-grains in the periplasm, we have the picture of the cytodal stage of evolution such as I have imagined it. It should be borne in mind that the ultimate granules of chromatin are probably in many cases ultra-microscopic; consequently they might appear to be dissolved in this cytoplasm when really existing as discrete particles.

In the life-cycles of Protozoa, especially of Rhizopods, it is not at all infrequent to find developmental phases which reproduce exactly the picture of the pseudo-moneral stage of evolution, phases in which the nucleus or nuclei have disappeared, having broken up into a number of chromatin-grains or chromidia scattered through the cytoplasm. We do not know as yet of any Protozoa, however, which remain permanently in the cytodal stage, that is to say, in which the chromatin-grains remain permanently in the scattered chromidial condition, without ever being concentrated and organized into true nuclei; but it is quite possible that some of the primitive organisms known as Proteomyxa will be found to exhibit this condition and to represent persistent Pseudo-monera or cytodes.

The next stage in evolution was the organization of the chromatin-grains (biococci) into a definite cell-nucleus. This is a process which can be observed actually taking place in many Protozoa in which "secondary" nuclei arise from chromidia. It seems not unreasonable to suppose that a detailed study of the manner in which secondary nuclei are formed in Protozoa will furnish us with a picture, or rather series of pictures, of the method in which the cell-nucleus arose in phylogeny. To judge from the data supplied by actual observation, the evolution of the nucleus, though uniform in principle, was sufficiently diversified in the details of the process. As one extreme we have the formation of a dense clump of small, separate

chromatin-grains, producing a granular nucleus of the type seen in Dinoflagellates, in Hæmogregarines, and in Diatoms. Amongst the chromatin-grains there may be present also one or more grains or masses of plastin forming true nucleoli. At the opposite extreme a clump of chromatin-grains becomes firmly welded together into a single mass in which the individual grains can no longer be distinguished, forming a so-called karyosome, consisting of a basis of plastin cementing or imbedding the chromatin-grains into a mass of homogeneous appearance. Whatever the type of nucleus formed, the concentration of the chromidia into nuclei does not necessarily involve all the chromidia, many of which may remain free in the cytoplasm.

In the chromidial condition the chromatin-grains scattered in the cytoplasm are lodged at the nodes of the alveolar framework.[24] Consequently a supporting framework of cytoplasmic origin, the foundation of the linin-framework, was probably a primary constituent of the cell-nucleus from the first. In many nuclei of the karyosomatic type it is very difficult to make out anything of the nature of a framework, which, however, in other cases is seen clearly as delicate strands radiating from the karyosome to the wall of the vacuole in which the karyosome is suspended. Probably such a framework is present in all cases, and each supporting strand is to be interpreted as the optical section of the partition between two protoplasmic alveoli.

With the formation of the nucleus the cytode or pseudomoneral stage has become a true cell of the simplest type, for which I propose the term *protocyte*. It is now the starting-point of an infinite series of further complications and elaborations in many directions. It is clearly

[24] Cf. Dobell, "Observations on the Life-History of Cienkowski's *Arachnula*," *Arch. Protistenkunde*, XXXI (1913), p. 322. The author finds that in *Arachnula* each nucleus arises from a single chromatin-grain, which grows to form a vesicular nucleus. Since the fully-formed nucleus contains numerous grains of chromatin, the original chromidiosome must multiply in this process.

impossible that I should do more than attempt to indicate in the most summary manner the various modifications of the cell-type of organism, since to deal with them conscientiously would require a treatise rather than an address, and, moreover, many such treatises exist already. The most conspicuous modifications of cell-structure are those affecting the periplasm, or, as we may now term it, the cytoplasm. In the first place, the cell as a whole takes various forms; primitively a little naked mass of protoplasm tending to assume a spherical form under the action of surface-tension when at rest, the cell-body may acquire the most diverse specific forms maintained either by the production of envelopes or various kinds of exoskeletal formations on the exterior of the protoplasmic body, or of supporting endoskeletal structures formed in the interior. The simple amœboid streaming movements become highly modified in various ways or replaced by special locomotor mechanisms or organs, flagella, cilia, etc., of various kinds. The internal alveolar cytoplasm develops fibrillæ and other structures of the most varied nature and function, contractile, skeletal, nervous, and so forth. The vacuole-system may be amplified and differentiated in various ways and the cytoplasm acquires manifold powers of internal or external secretion. And finally the cytoplasm contains enclosures of the most varied kind, some of them metaplastic products of the anabolic or catabolic activity essential to the maintenance of life, others of the nature of special cell-organs performing definite functions, such as centrosomes, plastids, chromatophores, etc., of various kinds.

With all the diverse modifications of the cytoplasmic cell-body the nucleus remains comparatively uniform. It may indeed vary infinitely in details of structure, but in principle it remains a concentration or aggregation of numerous grains of chromatin supported on some sort of framework over which the grains are scattered or clumped in various ways, supplemented usually by plastin or nucleolar substance either as a cementing ground-sub-

stance or as discrete grains, and the whole marked off sharply from the surrounding cytoplasm, with or without a definite limiting membrane. There is, however, one point in which the nucleus exhibits a progressive evolution of the most important kind. I refer to the gradual elaboration and perfection of the reproductive mechanism, the process whereby, when the cell reproduces itself by fission, the chromatin-elements are distributed between the two daughter-cells.

The chromatin-constituents of the cell are regarded, on the view maintained here, as a number of minute granules, each representing a primitive independent living individual or biococcus. To each such granule must be attributed the fundamental properties of living organisms in general; in the first place metabolism, expressed in continual molecular change, in assimilation and in growth, with consequent reproduction; in the second place specific individuality. As the result of the first of these properties the chromatin-granules, often perhaps ultra-microscopic, may be larger or smaller at different times, and they multiply by dividing each into two daughter-granules. As a result of the second property, chromatin-granules in one and the same cell may exhibit qualitative differences and may diverge widely from one another in their reactions and effects on the vital activities of the cell. The chromatin-granules may be either in the form of scattered chromidia or lodged in a definite nucleus. When in the former condition, I have proposed the term chromidiosome[25] for the ultimate chromatinic individual unit; on the other hand, the term chromiole is commonly in use for the minute chromatin-grains of the nucleus. The terms chromidiosome and chromiole distinguish merely between the situation in the cell, extranuclear or intranuclear, of the individual chromatin-grain or biococcus.

[25] "*Introduction of the Study of the Protozoa,*" Arnold, 1912, p. 65.

SHORTER ARTICLES AND DISCUSSION

INHERITANCE OF CONGENITAL CATARACT

CATARACT is the opacity of the eye caused by a faulty formation of the lens. Certain forms of cataract are congenital and hereditary. Other forms which appear later in life may either be hereditary or due to pathological causes.

In the normal eye the delicate fibers which go to make up the lens are glued together along their sides and at their ends where they unite in lines radiating from the poles of the lens to form a completely transparent body. Anything which prevents the perfect conjunction of these fibers causes a defect in the transparency of the lens. This imperfection has been compared by Harman (2) to the white spots in the finger nail, caused by slight injuries to the nail bed, and he has shown it to be correlated with faulty formation of the dental enamel.

There are various causes for the inhibition of proper lens development, and these give rise to different forms of cataract. Only those forms have been considered here which are congenital.

The most common form of congenital cataract is the lamellar, perinuclear or zonular cataract. This manifests itself as a dark circular disk with the density increasing from the center to the perimeter, forming characteristic zones. These zones are flecked by small wedge-shaped dashes arranged regularly in a spoke-fashion about the disk. The disk is located between the nucleus of the lens and the cortex; and is caused by a thickening of the layers at that place.

Discoid cataract is a slight form of the lamellar, less than 4 mm. in diameter, and located at the posterior pole of the lens. The opacity is uniform throughout, but is not easily visible. (It is sometimes confused with anterior polar cataract, of which the origin is not definitely known, but which is not congenital.)

Coralliform or axial cataract, *cataracta fusiformis*, is an opaque line running through the lens from anterior to posterior pole with a spindle-shaped swelling towards the center of the lens.

Anterior and posterior cortical cataract, *cataracta corticallis*, where the opacity takes a more or less geometrical outline,

cataracta punctata, formed by minute white dots scattered uniformly through the lens or grouped in the anterior cortical layers, and other forms of circumscribed, stationary, lenticular opacities which though rare are known to be congenital (3), have also been used in the compilations given here.

Senile cataract, which also seems to be hereditary, and those forms of cataract arising from lesions, diseases of the eyeball, and certain general diseases such as cholera and tetany, have been omitted.

Although congenital and other forms of cataract have long been considered by the medical profession to be influenced by heredity, no definite analysis was made until 1905, when the first paper by Nettleship (1) appeared. Nettleship's data have been the basis of Bateson's (9) conclusion that the abnormality is inherited as a dominant character. Bateson acknowledges that normal parents have produced abnormal children, but these cases he explains as either *origin de novo,* or due to faulty classification of the parents, who in reality may have been slightly affected with cataract.

Davenport (3) has followed Bateson's conclusion in regard to the inheritance of cataract, and makes the eugenic recommendation that unaffected parents from affected stock may marry without fear of producing abnormal children.

In the "Treasury of Human Inheritance" Harman (2) gives one hundred genealogical tables dealing with congenital cataract. Each table represents two or more generations with a detailed account of the condition of each individual in regard to congenital defects of the eye. The data used in this paper have been taken from these tables. Only those families are used in which there is no doubt as to the condition of the parents or the children in respect to the abnormality, and where there is no question as to the total number of children in each family. After discarding all the doubtful cases, and picking a sibship with its parents from the table as a family, there is left a total of one hundred and twenty-five families which are classified into three different categories, as follows: (*A*) both parents normal with at least one abnormal child; (*B*) one parent normal, the other affected with some form of congenital cataract, with at least one abnormal child; (*C*) both parents abnormal, giving only abnormal children.

There are 31 families with both parents normal which give some abnormal children. In a total of 153 children from these

families, 61 are affected with cataract. This suggests that the character is more likely to be inherited as a recessive than as a dominant. Surely it is not possible to explain so many cases as *origin de novo* or as due to faulty classification of the parents.

In the second category given above (*B*), where one of the parents is affected and the other normal, the number of defective children would be expected to be approximately the same whether the character was inherited as a dominant or a recessive.

If the abnormality is considered as a recessive character, the ratio of 61 affected to 92 unaffected, already spoken of as having been obtained in the first category of families, shows an excess of recessives over the simple Mendelian expectancy for a monohybrid. This is to be expected since the criterion for including any family in the tabulation is the production of at least one abnormal child. In families with a small number of children it is probable that in some cases only normal children are produced in matings of heterozygote by heterozygote which should give, on the average, one fourth recessive. The observed results must then be compared to a modified Mendelian ratio which will allow for the omission of all-normal progenies. Such ratios have been calculated by Apert (4) and by Wright (5). The expected proportions given here are calculated according to the method given by the latter.

The proportion of recessives varies according to the number of children in the family and ranges, for a three-to-one ratio, from 100 per cent. in families with one child to very nearly 25 per cent. in families with fifteen children. The proportion is calculated from the formula

$$X = \frac{1}{4[1 - (\frac{3}{4})^N]},$$

where N is the number of children. Since the criterion for including any family is the production of one abnormal child, all families with one child must have 100 per cent. abnormal children. The proportion decreases, according to the law of chance, as the number of children in the family increases, finally reaching 25 per cent. as the number of children becomes large.

Table I compares the results obtained with the theoretical expectancy, worked out according to this method.

The method used for testing the agreement of the observed result with the theoretical is the one given by Pearson (6) and Elderton (7). It was originally used to test various series of bio-

TABLE I

NORMAL × NORMAL (BOTH HETEROZYGOUS — Nn × Nn)

Size of Family N	Number of Families	Total No. of Children	Calculated Proportion Recessive X	Calculated Number of Recessives C	Observed Number of Recessives O	$O-C$	$\frac{(O-C)^2}{C}$
1	2	2	1.0000	2.00	2	.00	.000
2	4	8	.5714	4.57	6	1.43	.447
3	6	18	.4324	7.78	13	5.22	3.502
4	5	20	.3657	7.31	8	.69	.065
5	0	0	.3278	.00	0	.00	.000
6	7	42	.3041	12.77	19	6.23	3.039
7	1	7	.2885	2.02	2	− .02	.000+
8	2	16	.2778	4.44	5	.56	.070
9	1	9	.2703	2.43	1	−1.43	.841
10	2	20	.2649	5.30	3	−2.30	.998
11	1	11	.2610	2.87	2	− .87	.263
	31	153		51.49	61		9.225

$N = 10.$
$P = .418$

	Normal	Abnormal	Per Cent. Abnormal
Observed	92	61	40
Calculated	102	51	33
Difference	10	10	7

logical measurements. Attention has been called to the application of this method of testing theoretical Mendelian ratios by Harris (8).

By calculation from the data given in Table I the measure of agreement, or "*P*," is .418. "*P*" is a value ranging from 0 to 1, proportional to the closeness with which the observed facts agree with the theoretical. In this case in four times out of ten, random samplings of similar data would give results deviating more widely from the theoretical. A possible explanation for this rather wide discrepancy will be given later.

The children from matings of normal by abnormal are given in Table II.

A total of 448 children from 90 families is used. 232 children out of the 448 are found to be defective, whereas 238 are expected according to the modified Mendelian ratio.

As before, the criterion for including any family in the tabulation is the production of at least one affected child. Only in this way can the matings of heterozygous dominants, *Nn* (normal), with homozygous recessives, *nn* (abnormal), be distin-

TABLE II

NORMAL × ABNORMAL (Nn × nn).

Size of Family N	Number of Families	Total No. of Children	Calculated Proportion Recessive X	Calculated Number of Recessives C	Observed Number of Recessives O	$O-C$	$\frac{(O-C)^2}{C}$
1	9	9	1.0000	9.00	9	.00	.000
2	11	22	.6667	14.67	20	5.33	1.936
3	10	30	.5714	17.14	19	1.86	.201
4	12	48	.5333	25.60	24	−1.60	.100
5	10	50	.5161	25.81	27	1.19	.054
6	8	48	.5080	24.38	23	−1.38	.078
7	11	77	.5039	38.80	40	1.20	.037
8	9	72	.5019	36.14	31	−5.14	.731
9	3	27	.5009	13.52	15	1.48	.162
10	3	30	.5005	15.02	9	−6.02	2.412
11	2	22	.5002	11.00	10	−1.00	.090
12	0	0	.5001	.00	0	.00	.000
13	1	13	.5001	6.50	5	−1.50	.346
	90	448		237.58	232		6.147

$N = 12.$
$P = .862$

	Normal	Abnormal	Per Cent. Abnormal
Observed	216	232	52
Calculated	210	238	53
Difference	6	6	1

guished from the matings of homozygous dominants, *NN*, with recessives. In the former matings approximately 50 per cent. of the children are expected to be abnormal. In the second case only normal children are expected, who should all be heterozygous for the abnormality. It is entirely possible in small families that matings which should give part abnormal offspring might give only normal children; but all these matings are excluded, for no distinction can be made between them and the more usual matings of abnormal with homozygous normal, also giving only normal children.

Having thus excluded part of the data, the modified ratio is calculated as before, except that in this case it applies to a one-to-one ratio, instead of a one-to-three. It ranges from 100 per cent. in families with one child, to very nearly 50 per cent. in families with fifteen children, and is calculated from the formula

$$X = \frac{1}{2[1 - (\frac{1}{2})^N]}.$$

The agreement of observation with expectancy is very close.

"*P*" having a value .862 means that, in nearly nine cases out of ten, other random samplings would deviate more widely from the theoretical.

The critical test as to whether or not congenital cataract can be considered as a simple recessive character lies in the matings of abnormal by abnormal. Families of this kind should have only abnormal children. Only three such matings are available. In two of these, five and two children, respectively, are the total numbers produced; and these are all abnormal. The other case is a doubtful one. Both parents are classified as having discoid cataract; one is given as seriously affected, the other only slightly so. Seven children are given for this mating: two are abnormal, and the others apparently normal. Three of these five died in infancy, so that their classification is doubtful, but there is no question as to the others. Assuming, as Bateson does in his cases, that there has been a faulty classification of the parents, that the parent given as slightly affected is not congenitally defective, but adventitiously so early in life; then this one discrepancy might be conveniently overlooked.

Another explanation may be found, however, in the fact that heterozygous individuals sometimes show the recessive character. Cases are known where a small percentage of heterozygous individuals show the recessive character, although the homozygous and heterozygous dominants are generally indistinguishable. If such is the case here, the slightly affected parent is heterozygous, and the occurrence of normal children is expected.

This assumption also helps to explain the deviation of observed results above the theoretical in Table I, and their deviation below the theoretical in Table II. If heterozygous individuals are sometimes classified as recessives, it would affect the classification of both the parents and the children. A few matings of abnormal (heterozygote showing the recessive character) by normal would be included in the category *B* which rightfully belong in category *A*. Thus matings giving a one-to-three ratio would be included among matings giving a one-to-one ratio. This would tend to reduce the observed results below the expected. A faulty classification of the children would tend to raise the results, but this would not be as strong a deviating factor as when influencing the parents and therefore a number of children. Thus the balance would tend slightly toward a decrease in the regular expectancy; a result which fits well with that obtained in Table II.

In the matings of normal by normal, there is, of course, no opportunity for this error to influence the classification of the parents, since abnormals, whether heterozygous or not, would not come here. But with the children, the number with the recessive character would be raised above the regular expectancy; a result which coincides with that in Table I.

If with more matings of abnormal by abnormal it is found that, with a few exceptions, only abnormal children are given, the evidence that cataract is a recessive character rather than a dominant will be fairly conclusive. It seems rather strange that congenital cataract manifesting itself, as it does, in such different ways, should be determined by a single unit factor. These things, however, must be explained in the simplest possible manner; an attempt to work it out with two or more factors would introduce great complications, and be practically impossible with the data as they have been gathered heretofore. The fact that a recessive character may not be recognized, for it occurs in mass data in a greater proportion than would be expected at first, should be noticed. Finally the approximation of the results obtained with those expected from the single unit factor form the best reason for its acceptance.

That certain geneticists should have laid down eugenic rules based on the inheritance of this character as a dominant is, at the very least, unfortunate. It is not only because of a mistake in the method of inheritance, but such rules should never be made until the exact hereditary processes are positively known, since such practises are likely, not only to bring discredit upon the science, but to injure people who endeavor to follow them in the regulations of their lives.

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HUXLEY AS A MUTATIONIST

ELSEWHERE I have pointed out that Galton[1] held with equal firmness to continuity and discontinuity in variation, and that the American horticulturist and botanist, Thomas Meehan,[2] held clear mutationist conceptions which he supported by accurate observations of variations in many plants. It seems worth while to add a note on the attitude of Huxley with regard to this question.

Whenever Huxley expressed himself on this matter he usually took occasion to say explicitly that he could see no reason why variations should not be discontinuous as well as continuous, and one of the few points on which he differed from Darwin was in ascribing greater significance to such marked changes. Several statements of his position in this matter are found in his volume of essays entitled *Darwiniana*.

Thus he says (p. 77):

> Mr. Darwin's position might, we think, have been even stronger than it is if he had not embarrassed himself with the aphorism "*natura non facit saltum*," which turns up so often in his pages. We believe, as we have said above, that Nature does make jumps now and then, and a recognition of the fact is of no small importance in disposing of many minor objections to the doctrine of transmutation.

Elsewhere (pp. 34, 404) Huxley refers to the well-known Ancon sheep, which originated from a single ram in the flock of a Massachusetts farmer named Seth Wight. The story of this breed of sheep is told in a letter from Col. David Humphreys to Sir Joseph Banks, then President of the Royal Society.[3] The farmer kept a flock of 15 ewes and one ram on the banks of the Charles River, at Dover, Mass., 16 miles from Boston. In 1791 a ram

[1] "Galton and Discontinuity in Variation," AMER. NAT., *48:* 697–699, 1914.

[2] "An Anticipatory Mutationist," AMER. NAT., *49:* 645–648, 1915.

[3] Humphreys, D., 1813, "On a New Variety in the Breeds of Sheep," *Phil. Trans. Roy. Soc.*, 1813: 88–95.

lamb was born having a short length of back and short bandy legs. Seeing an advantage in such an animal owing to its inability to jump fences, it was bred to the flock, the original ram being killed. The first year thereafter two lambs had the peculiarities of their father, and in following years a number more Ancon lambs were produced. The latter when bred together always, with one questionable exception, produced Ancons.

Hence the character was evidently a recessive, having originated from the normal through a negative variation or mutation, presumably in one germ cell. This being the case, the variation must have been carried in a latent or recessive condition for a certain number of generations until inbreeding brought it out in a homozygous form. The original ram which was killed must have been heterozygous for this character, also one at least of the ewes and probably more; for one such heterozygous ewe was necessary to produce the original Ancon ram, and the two Ancons which appeared next year in the back-cross not improbably came from different mothers. It is therefore impossible to say just how long this condition may have been handed on in a "latent" condition before inbreeding brought it out.

With few exceptions, the Ancons showed alternative inheritance when crossed with normal sheep, and (l. c., p. 90).

> Frequent instances have happened where common ewes have had twins by Ancon rams, when one exhibited the complete marks of features of the ewe; the other of the ram.

Incidentally this shows that such twins came from separate ova.

In a flock the Ancon sheep tended to keep together and separate from the normal members of the flock. The breed seems to have attained some popularity, but their flabby subscapular muscles, infirm construction, loose joints, crooked forelegs and awkward gait, while preventing them from jumping fences made them difficult to drive to market. Butchers also found the carcasses smaller and less saleable, so that they were soon supplanted after the introduction of the Merino. They were already scarce in 1813 and afterwards became extinct.

Huxley remarks regarding this case:

> Varieties then arise we know not why; and it is more than probable that the majority of varieties have arisen in this "spontaneous" manner, though we are, of course, far from denying that they may be traced, in some cases, to distinct external influences. . . . But however they may have arisen, what especially interests us at present is, to remark

that, once in existence, many varieties obey the fundamental law of reproduction that like tends to produce like; and their offspring exemplify it by tending to exhibit the same deviation from the parental stock as themselves.

After further discussing the case, Huxley remarks (Op. cit. p. 39):

> Here, then, is a remarkable and well-established instance, not only of a very distinct race being established *per saltum*, but of that race breeding "true" at once, and showing no mixed forms, even when crossed with another breed.

Réaumur's case of a Maltese couple having a hexadactylous son, three of whose four children were again hexadactylous, also comes in for Huxley's comment (p. 35 ff.). The following dicta on the subject of variation, from the same volume, are also worth quoting:

> Indeed we have always thought that Mr. Darwin unnecessarily hampered himself by adhering so strictly to his favourite "Nature non facit saltum." We greatly suspect that she does make considerable jumps in the way of variation now and then, and that these saltations give rise to some of the gaps which appear to exist in the series of known forms (p. 97).
>
> I apprehend that the foundation of the theory of natural selection is the fact that living bodies tend incessantly to vary. This variation is neither indefinite, nor fortuituous, nor does it take place in all directions, in the strict sense of these words. . . . A whale does not tend to vary in the direction of producing feathers, nor a bird in the direction of developing whalebone (p. 181).
>
> The importance of natural selection will not be impaired even if further inquiries should prove that variability is definite, and is determined in certain directions rather than in others, by conditions inherent in that which varies. It is quite conceivable that every species tends to produce varieties of a limited number and kind, and that the effect of natural selection is to favour the development of some of these, while it opposes the development of others along their predetermined lines of modification (p. 223).

From these and similar statements it appears evident that were Huxley living to-day he could scarcely escape being classed as a mutationist.

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